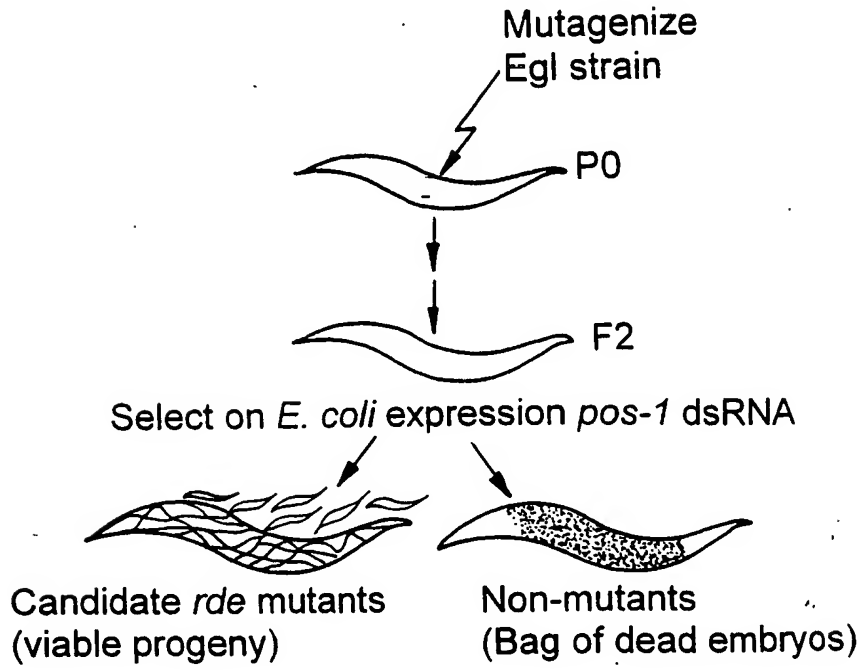
Mutagenize
Egl strain
P0
F2
Select on E. coli expression pos-1 dsRNA
Candidate rde mutants
(viable progeny)
Non-mutants
(Bag of dead embryos)

FIG. 1A

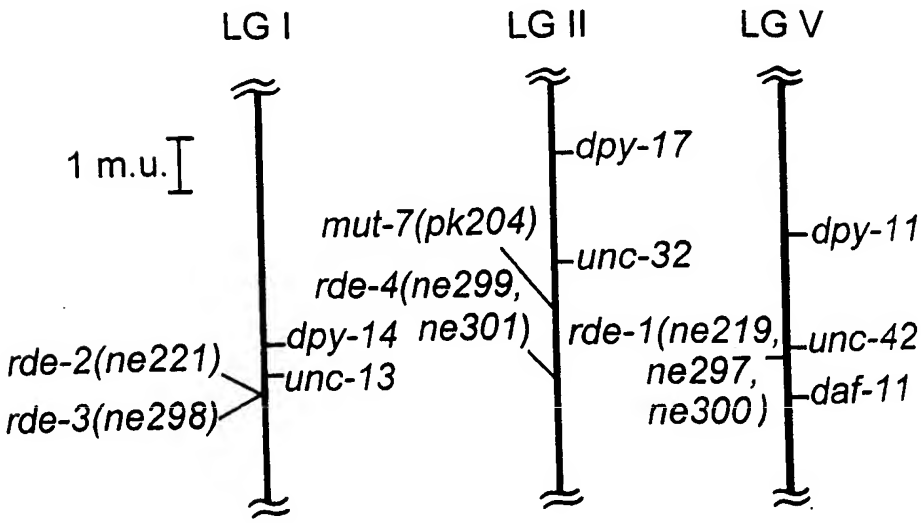
LG I
LG II
LG V
1 m.u.
dpy-17
mut-7(pk204)
unc-32
rde-4(ne299,
ne301)
dpy-11
rde-2(ne221)
dpy-14
unc-13
rde-1(ne219,
ne297,
ne300)
unc-42
daf-11
rde-3(ne298)

FIG. 1B

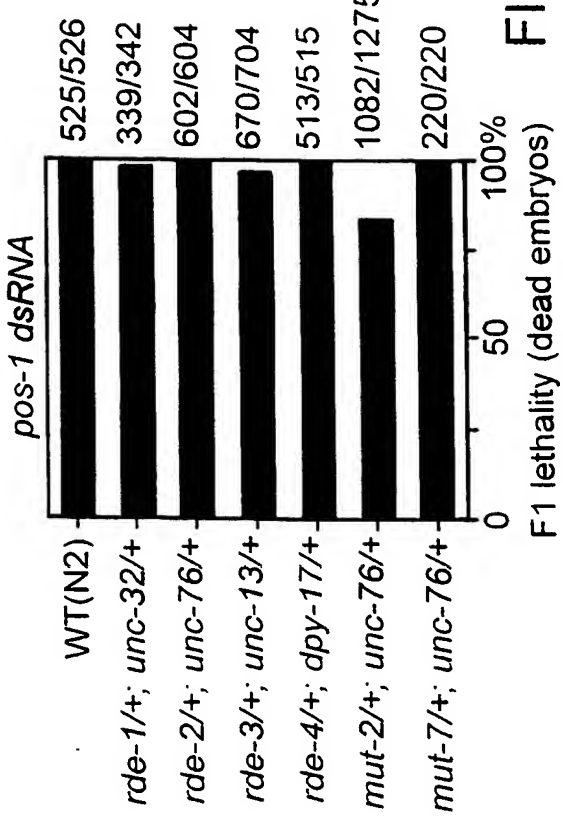
pos-1 dsRNA
WT(N2)
rde-1/+; unc-32/+
rde-2/+; unc-76/+
rde-3/+; unc-13/+
rde-4/+; dpy-17/+
mut-2/+; unc-76/+
mut-7/+; unc-76/+
525/526
339/342
602/604
670/704
513/515
1082/1275
220/220
0
50
100%
F1 lethality (dead embryos)

FIG. 2A

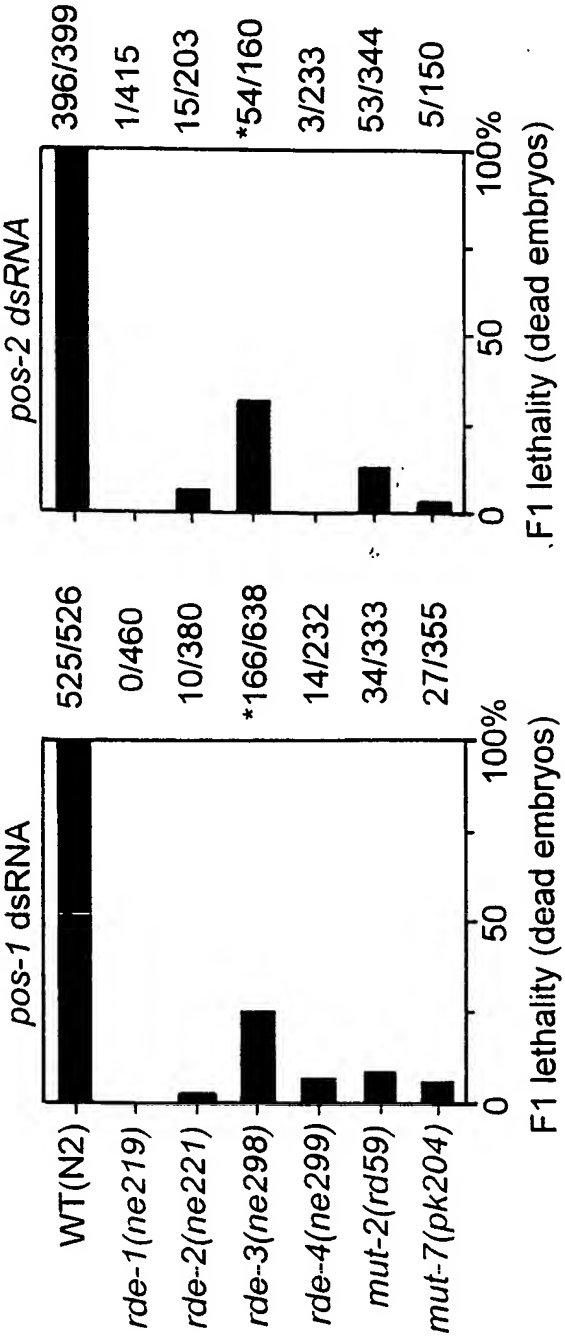
pos-1 dsRNA
WT(N2)
rde-1(ne219)
rde-2(ne221)
rde-3(ne298)
rde-4(ne299)
mut-2(rd59)
mut-7(pk204)
525/526
0/460
10/380
*166/638
14/232
34/333
27/355
pos-2 dsRNA
396/399
1/415
15/203
*54/160
3/233
53/344
5/150
0
50
100%
F1 lethality (dead embryos)

FIG. 2B

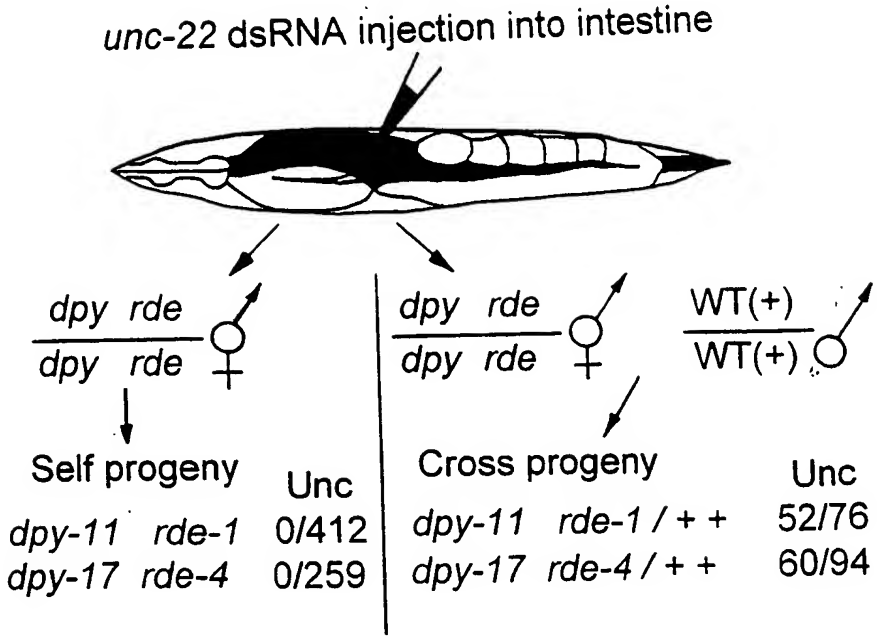
unc-22 dsRNA injection into intestine
dpy rde
dpy rde
Self progeny
Unc
dpy-11 rde-1 0/412
dpy-17 rde-4 0/259
dpy rde
dpy rde
WT(+)
WT(+)
Cross progeny
Unc
dpy-11 rde-1 / + + 52/76
dpy-17 rde-4 / + + 60/94

FIG. 3

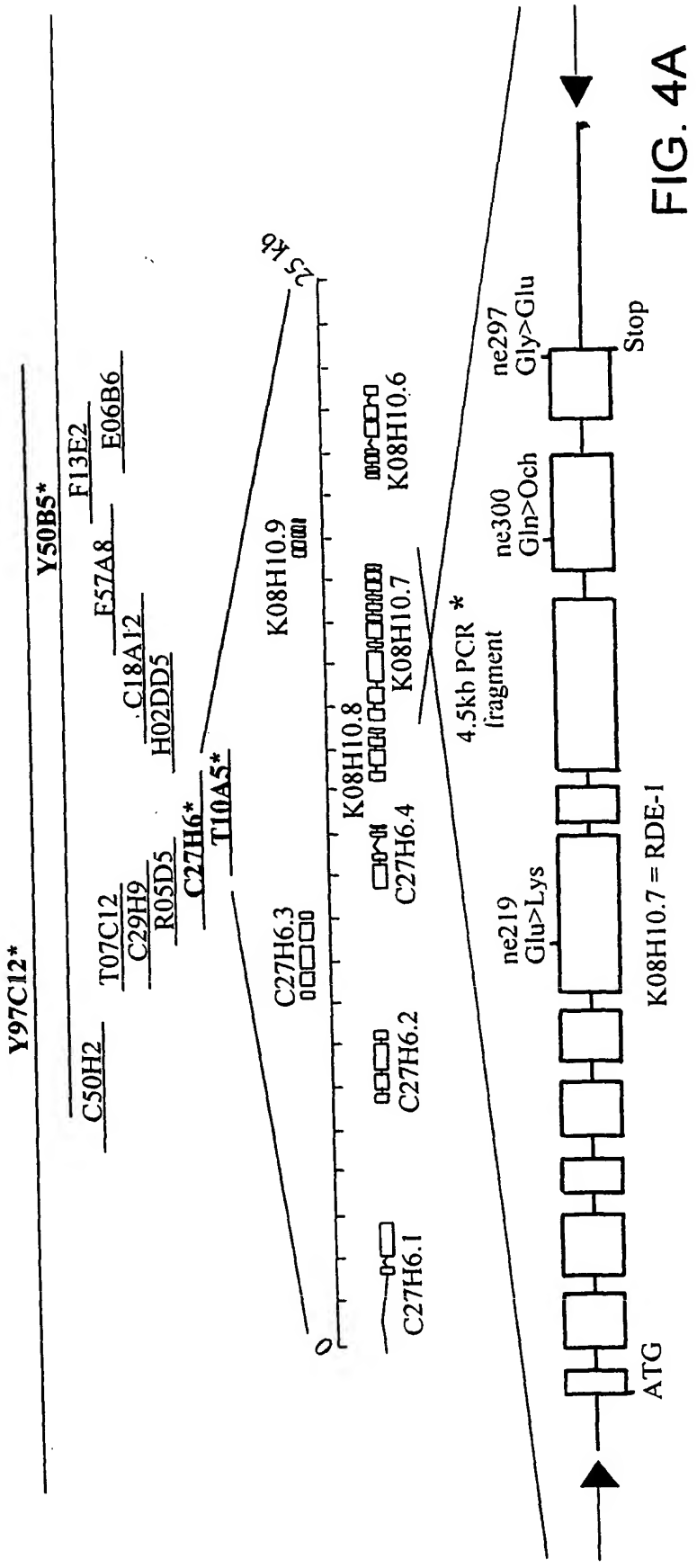
Y97C12*
Y50B5*
C50H2
T07C12
C29H9
R05D5
C27H6*
T10A5*
F13E2
E06B6
E57A8
C18A12
H02DD5
25 kb
0
K08H10.9
K08H10.8
K08H10.7
K08H10.6
C27H6.3
C27H6.4
C27H6.2
C27H6.1
4.5kb PCR *
fragment
ne219
Glu>Lys
ne300
Gln>Och
ne297
Gly>Glu
Stop
ATG
K08H10.7 = RDE-1

FIG. 4A

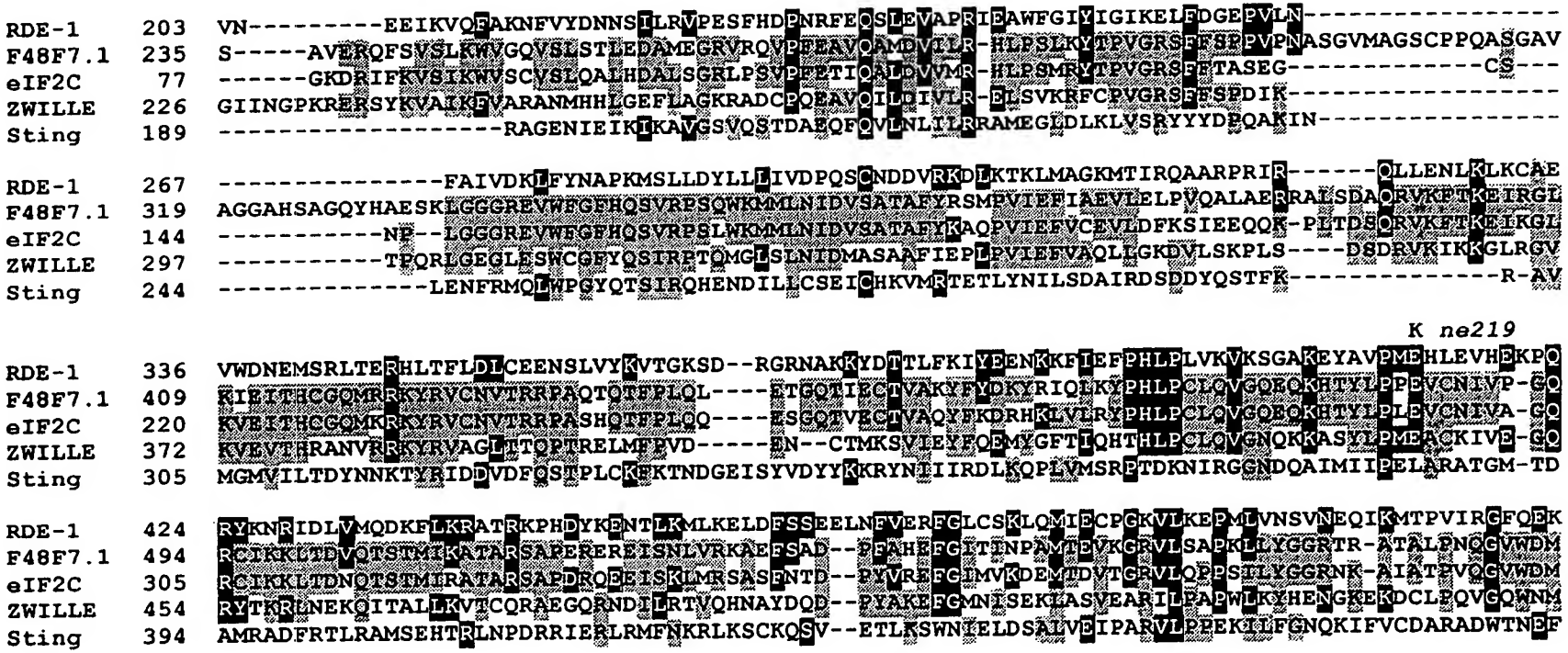
RDE-1 203 VN---------EEIKVQEAKNFVYDNNSILRVPESFHDPNRFEQSLEVAPRIEAWFGIYIGIKELEDGEPVLN----------
F48F7.1 235 S-----AVERQFSVSLKWVGQVSLSTLEDAMEGRVRQVPEEAVQAMDVILR-HLPSLKYTPVGRSFFSPPVPNASGVMAGSCPPQASGAV
eIF2C 77 ------GKDRIFKVSIKNVSCVSLQALHDALSGRLPSVPFETIQALDVVMR-HLPSMRYTPVGRSFFTASEG----------CS---
ZWILLE 226 GIINGPKRERSYKVAIKEVARANMHHLGEFLAGKRADCPQEAVQILQIVLR-ELSVKRFCPVGRSFFSPDIK----------
Sting 189 ----------RAGENIEIKIKAVGSVQSTDAEQFQVLNLILRRAMEGLDLKLVSRYYYDPQAKIN----------
RDE-1 267 ----------FAIVDKLFYNAPKMSLLDYLLLIVDPQSCNDDVRKDLKTKLMAGKMTIRQAARPRIR------QLLENLKLKCAE
F48F7.1 319 AGGAHSAGQYHAESKLGGGREVWEGFHQSVRPSQWKMMLNIDVSATAFYRSMPVIEFIAEVLELPVQALAERRALSDAQRVKFTKEIRGL
eIF2C 144 ----------NF--LGGGREVWEGFHQSVRPSLWKMMLNIDVSATAFYKAQPVIEFVCEVLDFKSIEEQQK-PLTDSQRVKFTKEIKGL
ZWILLE 297 ----------TPQRLGEGLESWCGFYQSIRPTQMGLSLNIDMASAAFIEPLPVIEFVAQLLGKDVLSKPLS----DSDRVKIKKGLRGV
Sting 244 ----------LENFRMQLWPGYQTSIRQHENDILLCSEICHKVMRTETLYNILSDAIRDSDDYQSTFK----------R-AV
K ne219
RDE-1 336 VWDNEMSRLTERHLTFLDLCEENSLVYKVTGKSD--RGRNAKKYDTTLFKIYEENKKFIEFPHLPLVKVKSGAKEYAVPMEHLEVHEKPQ
F48F7.1 409 KIEITHCGQMRRKYRVCNVTRRPAQTQTFPLQL----ETGQTIECTVAKYFYDKYRIQLKYPHLPCLQVGQEQKHTYLPPEVCNIVP-GQ
eIF2C 220 KVEITHCGQMKRKYRVCNVTRRPASHQTFPLQQ----ESGQTVECTVAQYFKDRHKLVLRYPHLPCLQVGQEQKHTYIPLEVCNIVA-GQ
ZWILLE 372 KVEVTHRANVRRKYRVAGLTTQPTRELMFPVD-----EN--CTMKSVIEYFQEMYGFTIQHTHLPCLQVGNQKKASYLPMEACKIVE-GQ
Sting 305 MGMVILTDYNNKTYRIDDVDFQSTPLCKFKTNDGEISYVDYYKKRYNIIIRDLKQPLVMSRPTDKNIRGGNDQAIMIIPELARATGM-TD
RDE-1 424 RYKNRIDLVMQDKFLKRATRKPHDYKENTLKMLKELDFSSEELNFVERFGLCSKLQMIECPGKVLKEPMLVNSVNEQIKMTPVIRGFQEK
F48F7.1 494 RCIKKLTDVQTSTMIKATARSAPERERETSNLVRKAEESAD--PEAHEFGITINPAMTEVKGRVLSAPKLLYGGRTR-ATALFNQGVWDM
eIF2C 305 RCIKKLTDNQTSTMIRATARSAPDRQEEISKLMRSASENTD--PYVREFGIMVKDEMTDVTGRVLQPPSILYGGRNK-AIATPVQGVWDM
ZWILLE 454 RYTKRLNEKQITALLKVTCQRAEGQRNDILRTVQHNAYDQD--PYAKEFGMNISEKLASVEARILPAPWLKYHENGKEKDCLFQVGQWNM
Sting 394 AMRADFRTLRAMSEHTRLNPDRRIERLRMFNKRLKSCKQSV--ETLKSWNIELDSALVEIPARVLPPEKILFGNQKIFVCDARADWTNEF

FIG. 4B-1

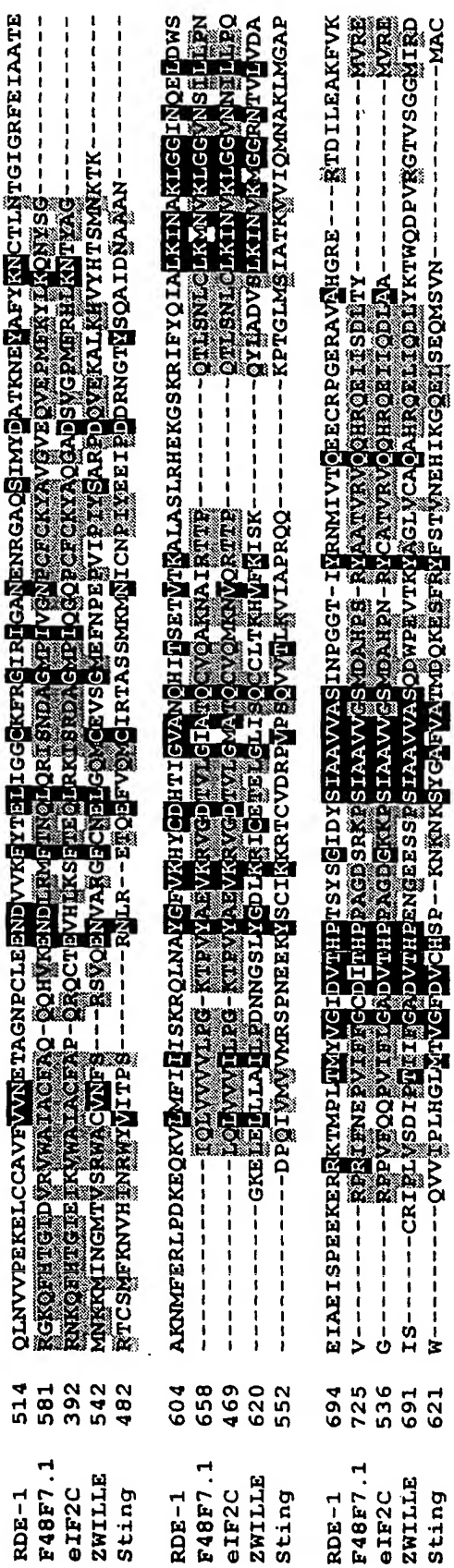

FIG. 4B-2

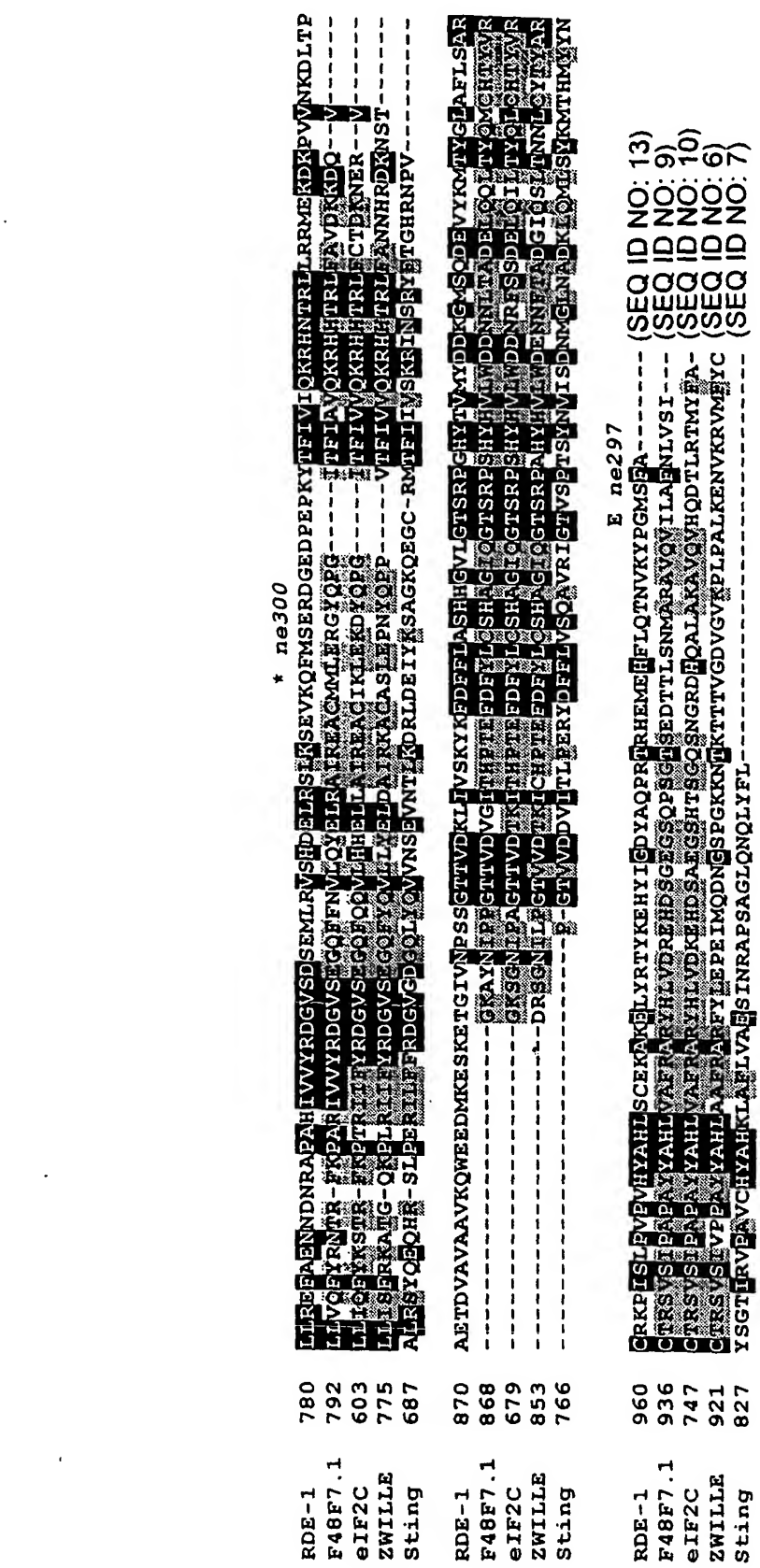
* ne300
E ne297
RDE-1
F48F7.1
eIF2C
ZWILLE
Sting
(SEQ ID NO: 13)
(SEQ ID NO: 9)
(SEQ ID NO: 10)
(SEQ ID NO: 6)
(SEQ ID NO: 7)

FIG. 4B-3

cagccacaaagtgatgaaacatgtcctcgaattttcccgaattggaaaaaggattttatcgtcattctctcgatccggta

tgatcaattattagcagctataagatatataagtttgatattaatattataggagatgaaatggcttgcgaggcccactg

gtaaatgcgacggcaaattctatgagaagaaagtacttcttttggtaaattggttcaagttctccagcaaaatttacgat

cgggaatactacgagtatgaagtgaaaatgacaaaggaagtattgaatagaaaaccaggaaaacctttcccaaaaaag

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aaaggatttggagaaaaaaatcttatacacaatgatacttacctatcgtaaaaaatttcacctgaactttagtcgagaaa

atccggaaaaagacgaagaagcgaatcggagttacaaattcctgaaggtttatgaaaaacacgcattataacaaacaa

ttagctttcagaatgttatgacccagaaagttcgctacgcgccttttgtgaacgaggagattaaagtgtgagttgcaata

ataataataatcacctcaactcatttatatattttaagacaattcgcgaaaaattttgtgtacgataataattcaat

FIG. 5A

tctgcgagttcctgaatcgtttcacgatccaaacagattcgaacaatcattagaagtagcaccaagaatcgaagcatggt

ttggaatttacattggaatcaaagaattgttcgatggtgaacctgtgctcaattttgcaagtaagtttgagaaactgcga

taaaaaatcatgtgatttttgttgaagttgtcgataaactattctacaatgcaccgaaaatgtctcttctggattatctt

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ttgtgactcaagaagaatgtcgtcccggtgagcgtgcagtggctcatggacgggaaagaacagatattttggaagcaaa

ttcgtgaaattgctcagagaattcgcagaagtgagttgtcttgagtatttaaaagatctctgggatttttaatttttttg

FIG. 5B

taaactttcagaacaacgacaatcgagcaccagcgcatattgtagtctatcgagacggagttagcgattcggagatgcta

cgtgttagtcatgatgagcttcgatctttaaaaagcgaagtaaaacaattcatgtcggaacgggatggagaagatccaga

gccgaagtacacgttcattgtgattcagaaaagacacaatacacgattgcttcgaagaatggaaaaagataagccagtg

tcaataaagatcttactcctgctgaaacagatgtcgctgttgctgctgttaaacaatgggaggaggatatgaaagaaagc

aaagaaactggaattgtgaacccatcatccggaacaactgtggataaacttatcgtttcgaaatacaaattcgatttttt

cttggcatctcatcatggtgtccttggtacatctcgtccaggacattacactgttatgtatgacgataaaggaatgagcc

aagatgaagtctatgtaagcgttttgaatagcagttagcgattttaggattttgtaatccgcatatagttattataaaaa

aatgtttcagaaaatgacctacggacttgcttttctctctgctagatgtcgaaaacccatctcgttgcctgttccggttc

attatgctcatttatcatgtgaaaaagcgaaagagctttatcgaacttacaaggaacattacatcggtgactatgcacag

ccacggactcgacacgaaatggaacattttctccaaactaacgtgaagtaccctggaatgtcgttcgcataacattttgc

aaaagtgtcgcccgtttcaatcaaatttttcaattgtagatattgtacttacttttttttaaagcccggtttcaaaaatt

cattccatgactaacgttttcataaattacttgaaattt (SEQ ID NO:1)

FIG. 5C

App No.: Not Yet Assigned
Inventor: Craig C. Mello et al.
Title: RNA INTERFERENCE PATHWAY GENES AS TOOLS FOR TARGETED GENETIC INTERFERENCE
Docket No.: UMG-052DV2

CAGCCACAAAGTGATGAAAC- 3' UTR

```
1/1                                                 31/11
ATG TCC TCG AAT TTT CCC GAA TTG GAA AAA GGA TTT TAT CGT CAT TCT CTC GAT CCG GAG
Met ser ser asn phe pro glu leu glu lys gly phe tyr arg his ser leu asp pro glu

61/21                                               91/31
ATG AAA TGG CTT GCG AGG CCC ACT GGT AAA TGC GAC GGC AAA TTC TAT GAG AAG AAA GTA
met lys trp leu ala arg pro thr gly lys cys asp gly lys phe tyr glu lys lys val

121/41                                              151/51
CTT CTT TTG GTA AAT TGG TTC AAG TTC TCC AGC AAA ATT TAC GAT CGG GAA TAC TAC GAG
leu leu leu val asn trp phe lys phe ser ser lys ile tyr asp arg glu tyr tyr glu

181/61                                              211/71
TAT GAA GTG AAA ATG ACA AAG GAA GTA TTG AAT AGA AAA CCA GGA AAA CCT TTC CCA AAA
tyr glu val lys met thr lys glu val leu asn arg lys pro gly lys pro phe pro lys

241/81                                              271/91
AAG ACA GAA ATT CCA ATT CCC GAT CGT GCA AAA CTC TTC TGG CAA CAT CTT CGG CAT GAG
lys thr glu ile pro ile pro asp arg ala lys leu phe trp gln his leu arg his glu

301/101                                             331/111
AAG AAG CAG ACA GAT TTT ATT CTC GAA GAC TAT GTT TTT GAT GAA AAG GAC ACT GTT TAT
lys lys gln thr asp phe ile leu glu asp tyr val phe asp glu lys asp thr val tyr

361/121                                             391/131
AGT GTT TGT CGA CTG AAC ACT GTC ACA TCA AAA ATG CTG GTT TCG GAG AAA GTA GTA AAA
ser val cys arg leu asn thr val thr ser lys met leu val ser glu lys val val lys

421/141                                             451/151
AAG GAT TCG GAG AAA AAA GAT GAA AAG GAT TTG GAG AAA AAA ATC TTA TAC ACA ATG ATA
lys asp ser glu lys lys asp glu lys asp leu glu lys lys ile leu tyr thr met ile

481/161                                             511/171
CTT ACC TAT CGT AAA AAA TTT CAC CTG AAC TTT AGT CGA GAA AAT CCG GAA AAA GAC GAA
leu thr tyr arg lys lys phe his leu asn phe ser arg glu asn pro glu lys asp glu

541/181                                             571/191
GAA GCG AAT CGG AGT TAC AAA TTC CTG AAG AAT GTT ATG ACC CAG AAA GTT CGC TAC GCG
glu ala asn arg ser tyr lys phe leu lys asn val met thr gln lys val arg tyr ala

601/201                                             631/211
CCT TTT GTG AAC GAG GAG ATT AAA GTA CAA TTC GCG AAA AAT TTT GTG TAC GAT AAT AAT
pro phe val asn glu glu ile lys val gln phe ala lys asn phe val tyr asp asn asn

661/221                                             691/231
TCA ATT CTG CGA GTT CCT GAA TCG TTT CAC GAT CCA AAC AGA TTC GAA CAA TCA TTA GAA
ser ile leu arg val pro glu ser phe his asp pro asn arg phe glu gln ser leu glu

721/241                                             751/251
```

FIG. 6A

GTA GCA CCA AGA ATC GAA GCA TGG TTT GGA ATT TAC ATT GGA ATC AAA GAA TTG TTC GAT
val ala pro arg ile glu ala trp phe gly ile tyr ile gly ile lys glu leu phe asp

781/261 811/271
GGT GAA CCT GTG CTC AAT TTT GCA ATT GTC GAT AAA CTA TTC TAC AAT GCA CCG AAA ATG
gly glu pro val leu asn phe ala ile val asp lys leu phe tyr asn ala pro lys met

841/281 871/291
TCT CTT CTG GAT TAT CTT CTC CTA ATT GTC GAC CCC CAG TCG TGT AAC GAT GAT GTA CGA
ser leu leu asp tyr leu leu leu ile val asp pro gln ser cys asn asp asp val arg

901/301 931/311
AAA GAT CTT AAA ACA AAA CTG ATG GCG GGA AAA ATG ACA ATC AGA CAA GCC GCG CGG CCA
lys asp leu lys thr lys leu met ala gly lys met thr ile arg gln ala ala arg pro

961/321 991/331
AGA ATT CGA CAA TTA TTG GAA AAT TTG AAG CTG AAA TGC GCA GAA GTT TGG GAT AAC GAA
arg ile arg gln leu leu glu asn leu lys leu lys cys ala glu val trp asp asn glu

1021/341 1051/351
ATG TCG AGA TTG ACA GAA CGA CAT CTG ACA TTT CTA GAT TTG TGC GAG GAA AAC TCT CTT
met ser arg leu thr glu arg his leu thr phe leu asp leu cys glu glu asn ser leu

1081/361 1111/371
GTT TAT AAA GTC ACT GGT AAA TCG GAC AGA GGA AGA AAT GCA AAA AAG TAC GAT ACT ACA
val tyr lys val thr gly lys ser asp arg gly arg asn ala lys lys tyr asp thr thr

1141/381 1171/391
TTG TTC AAA ATC TAT GAG GAA AAC AAA AAG TTC ATT GAG TTT CCC CAC CTA CCA CTA GTC
leu phe lys ile tyr glu glu asn lys lys phe ile glu phe pro his leu pro leu val

1201/401 1231/411
AAA GTT AAA AGT GGA GCA AAA GAA TAC GCT GTA CCA ATG GAA CAT CTT GAA GTT CAT GAG
lys val lys ser gly ala lys glu tyr ala val pro met glu his leu glu val his glu

1261/421 1291/431
AAG CCA CAA AGA TAC AAG AAT CGA ATT GAT CTG GTG ATG CAA GAC AAG TTT CTA AAG CGA
lys pro gln arg tyr lys asn arg ile asp leu val met gln asp lys phe leu lys arg

1321/441 1351/451
GCT ACA CGA AAA CCT CAC GAC TAC AAA GAA AAT ACC CTA AAA ATG CTG AAA GAA TTG GAT
ala thr arg lys pro his asp tyr lys glu asn thr leu lys met leu lys glu leu asp

1381/461 1411/471
TTC TCT TCT GAA GAG CTA AAT TTT GTT GAA AGA TTT GGA TTA TGC TCC AAA CTT CAG ATG
phe ser ser glu glu leu asn phe val glu arg phe gly leu cys ser lys leu gln met

1441/481 1471/491
ATC GAA TGT CCA GGA AAG GTT TTG AAA GAG CCA ATG CTT GTG AAT AGT GTA AAT GAA CAA
ile glu cys pro gly lys val leu lys glu pro met leu val asn ser val asn glu gln

1501/501 1531/511
ATT AAA ATG ACA CCA GTG ATT CGT GGA TTT CAA GAA AAA CAA TTG AAT GTG GTT CCC GAA
ile lys met thr pro val ile arg gly phe gln glu lys gln leu asn val val pro glu

FIG. 6B

1561/521
AAA GAA CTT TGC TGT GCT GTT TTT GTA GTC AAC GAA ACA GCG GGA AAT CCA TGC TTA GAA
lys glu leu cys cys ala val phe val val asn glu thr ala gly asn pro cys leu glu

1621/541
GAG AAC GAC GTT GTT AAG TTC TAC ACC GAA CTA ATT GGT GGT TGC AAG TTC CGT GGA ATA
glu asn asp val val lys phe tyr thr glu leu ile gly gly cys lys phe arg gly ile

1681/561
CGA ATT GGT GCC AAT GAA AAC AGA GGA GCG CAA TCT ATT ATG TAC GAC GCG ACG AAA AAT
arg ile gly ala asn glu asn arg gly ala gln ser ile met tyr asp ala thr lys asn

1741/581
GAA TAT GCC TTC TAC AAA AAT TGT ACA CTA AAT ACC GGA ATC GGT AGA TTT GAA ATA GCC
glu tyr ala phe tyr lys asn cys thr leu asn thr gly ile gly arg phe glu ile ala

1801/601
GCA ACA GAA GCG AAG AAT ATG TTT GAA CGT CTT CCC GAT AAA GAA CAA AAA GTC TTA ATG
ala thr glu ala lys asn met phe glu arg leu pro asp lys glu gln lys val leu met

1861/621
TTC ATT ATC ATT TCC AAA CGA CAA CTG AAT GCT TAC GGT TTT GTG AAA CAT TAT TGC GAT
phe ile ile ile ser lys arg gln leu asn ala tyr gly phe val lys his tyr cys asp

1921/641
CAC ACC ATC GGT GTA GCT AAT CAG CAT ATT ACT TCT GAA ACA GTC ACA AAA GCT TTG GCA
his thr ile gly val ala asn gln his ile thr ser glu thr val thr lys ala leu ala

1981/661
TCA CTA AGG CAC GAG AAA GGA TCA AAA CGA ATT TTC TAT CAA ATT GCA TTG AAA ATC AAC
ser leu arg his glu lys gly ser lys arg ile phe tyr gln ile ala leu lys ile asn

2041/681
GCG AAA TTA GGA GGT ATT AAC CAG GAG CTT GAC TGG TCA GAA ATT GCA GAA ATA TCA CCA
ala lys leu gly gly ile asn gln glu leu asp trp ser glu ile ala glu ile ser pro

2101/701
GAA GAA AAA GAA AGA CGG AAA ACA ATG CCA TTA ACT ATG TAT GTT GGA ATT GAT GTA ACT
glu glu lys glu arg arg lys thr met pro leu thr met tyr val gly ile asp val thr

2161/721
CAT CCA ACC TCC TAC AGT GGA ATT GAT TAT TCT ATA GCG GCT GTA GTA GCG AGT ATC AAT
his pro thr ser tyr ser gly ile asp tyr ser ile ala ala val val ala ser ile asn

2221/741
CCA GGT GGA ACT ATC TAT CGA AAT ATG ATT GTG ACT CAA GAA GAA TGT CGT CCC GGT GAG
pro gly gly thr ile tyr arg asn met ile val thr gln glu glu cys arg pro gly glu

2281/761
CGT GCA GTG GCT CAT GGA CGG GAA AGA ACA GAT ATT TTG GAA GCA AAG TTC GTG AAA TTG
arg ala val ala his gly arg glu arg thr asp ile leu glu ala lys phe val lys leu

2341/781
CTC AGA GAA TTC GCA GAA AAC AAC GAC AAT CGA GCA CCA GCG CAT ATT GTA GTC TAT CGA
leu arg glu phe ala glu asn asn asp asn arg ala pro ala his ile val val tyr arg

FIG. 6C

2401/801 2431/811
GAC GGA GTT AGC GAT TCG GAG ATG CTA CGT GTT AGT CAT GAT GAG CTT CGA TCT TTA AAA
asp gly val ser asp ser glu met leu arg val ser his asp glu leu arg ser leu lys

2461/821 2491/831
AGC GAA GTA AAA CAA TTC ATG TCG GAA CGG GAT GGA GAA GAT CCA GAG CCG AAG TAC ACG
ser glu val lys gln phe met ser glu arg asp gly glu asp pro glu pro lys tyr thr
2521/841 2551/851
TTC ATT GTG ATT CAG AAA AGA CAC AAT ACA CGA TTG CTT CGA AGA ATG GAA AAA GAT AAG
phe ile val ile gln lys arg his asn thr arg leu leu arg arg met glu lys asp lys

2581/861 2611/871
CCA GTG GTC AAT AAA GAT CTT ACT CCT GCT GAA ACA GAT GTC GCT GTT GCT GCT GTT AAA
pro val val asn lys asp leu thr pro ala glu thr asp val ala val ala ala val lys

2641/881 2671/891
CAA TGG GAG GAG GAT ATG AAA GAA AGC AAA GAA ACT GGA ATT GTG AAC CCA TCA TCC GGA
gln trp glu glu asp met lys glu ser lys glu thr gly ile val asn pro ser ser gly

2701/901 2731/911
ACA ACT GTG GAT AAA CTT ATC GTT TCG AAA TAC AAA TTC GAT TTT TTC TTG GCA TCT CAT
thr thr val asp lys leu ile val ser lys tyr lys phe asp phe phe leu ala ser his

2761/921 2791/931
CAT GGT GTC CTT GGT ACA TCT CGT CCA GGA CAT TAC ACT GTT ATG TAT GAC GAT AAA GGA
his gly val leu gly thr ser arg pro gly his tyr thr val met tyr asp asp lys gly

2821/941 2851/951
ATG AGC CAA GAT GAA GTC TAT AAA ATG ACC TAC GGA CTT GCT TTT CTC TCT GCT AGA TGT
met ser gln asp glu val tyr lys met thr tyr gly leu ala phe leu ser ala arg cys

2881/961 2911/971
CGA AAA CCC ATC TCG TTG CCT GTT CCG GTT CAT TAT GCT CAT TTA TCA TGT GAA AAA GCG
arg lys pro ile ser leu pro val pro val his tyr ala his leu ser cys glu lys ala

2941/981 2971/991
AAA GAG CTT TAT CGA ACT TAC AAG GAA CAT TAC ATC GGT GAC TAT GCA CAG CCA CGG ACT
lys glu leu tyr arg thr tyr lys glu his tyr ile gly asp tyr ala gln pro arg thr

3001/1001 3031/1011
CGA CAC GAA ATG GAA CAT TTT CTC CAA ACT AAC GTG AAG TAC CCT GGA ATG TCG TTC GCA
arg his glu met glu his phe leu gln thr asn val lys tyr pro gly met ser phe ala

3061/1021 3091/1031
TAA CAT TTT GCA AAA GTG TCG CCC GTT TCA ATC AAA TTT TTC AAT TGT AGA TAT TGT ACT
CCH (SEQ ID NO:3)

3121/1041 3151/1051
TAC TTT TTT TTA AAG CCC GGT TTC AAA AAT TCA TTC CAT GAC TAA CGT TTT CAT AAA TTA

3181/1061
CTT GAA ATT TAA AAA AAA AAA AAA AAA (SEQ ID NO:2)

FIG. 6D

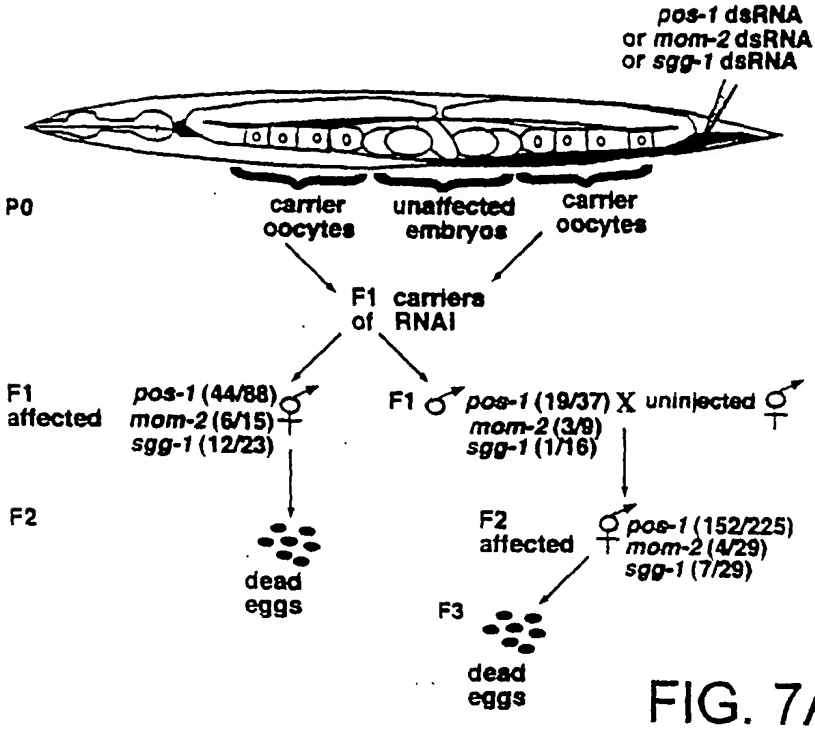

pos-1 dsRNA
or mom-2 dsRNA
or sgg-1 dsRNA
P0
carrier oocytes
unaffected embryos
carrier oocytes
F1 carriers of RNAi
F1 affected
pos-1 (44/88)
mom-2 (6/15)
sgg-1 (12/23)
F1
pos-1 (19/37) X uninjected
mom-2 (3/9)
sgg-1 (1/16)
F2
dead eggs
F2 affected
pos-1 (152/225)
mom-2 (4/29)
sgg-1 (7/29)
F3
dead eggs

FIG. 7A

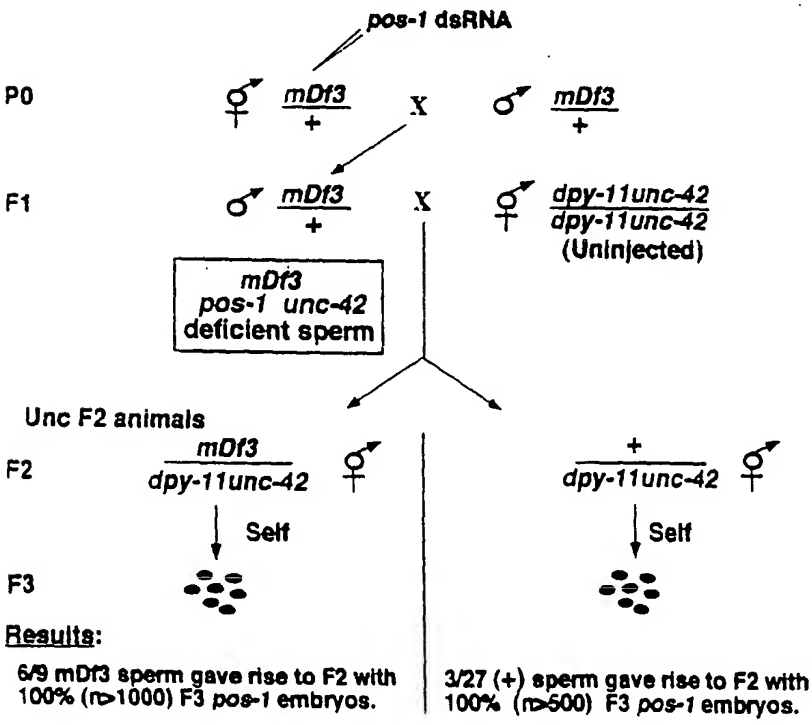

pos-1 dsRNA
P0
mDf3 / + X mDf3 / +
F1
mDf3 / + X dpy-11unc-42 / dpy-11unc-42 (Uninjected)
mDf3 pos-1 unc-42 deficient sperm
Unc F2 animals
F2
mDf3 / dpy-11unc-42
Self
F3
+ / dpy-11unc-42
Self
Results:
6/9 mDf3 sperm gave rise to F2 with 100% (n>1000) F3 pos-1 embryos.
3/27 (+) sperm gave rise to F2 with 100% (n>500) F3 pos-1 embryos.

FIG. 7B

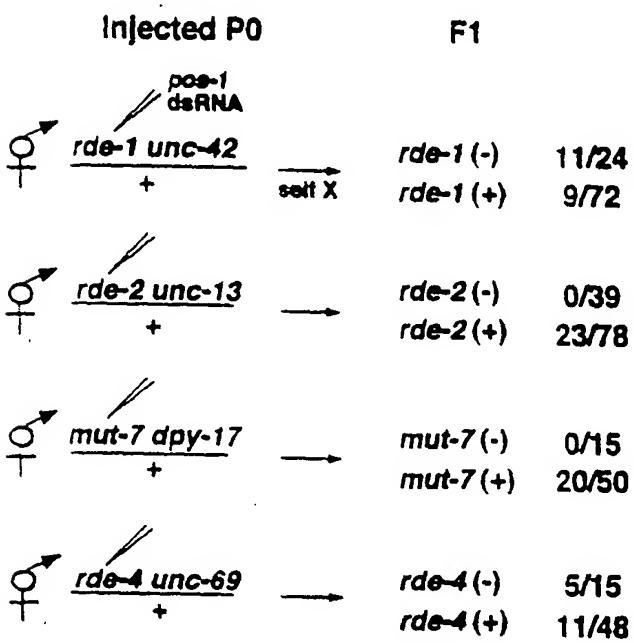

Injected P0
F1
pos-1 dsRNA
rde-1 unc-42
+
self X
rde-1 (-) 11/24
rde-1 (+) 9/72
rde-2 unc-13
+
rde-2 (-) 0/39
rde-2 (+) 23/78
mut-7 dpy-17
+
mut-7 (-) 0/15
mut-7 (+) 20/50
rde-4 unc-69
+
rde-4 (-) 5/15
rde-4 (+) 11/48

FIG. 8A

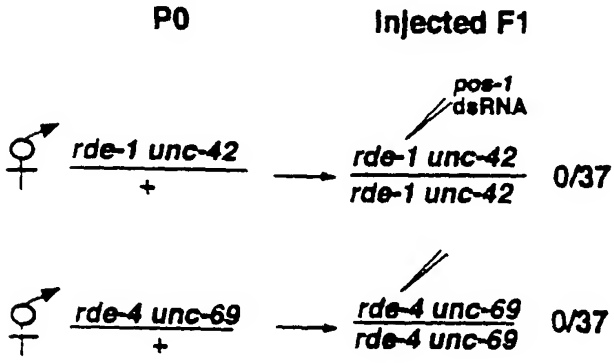
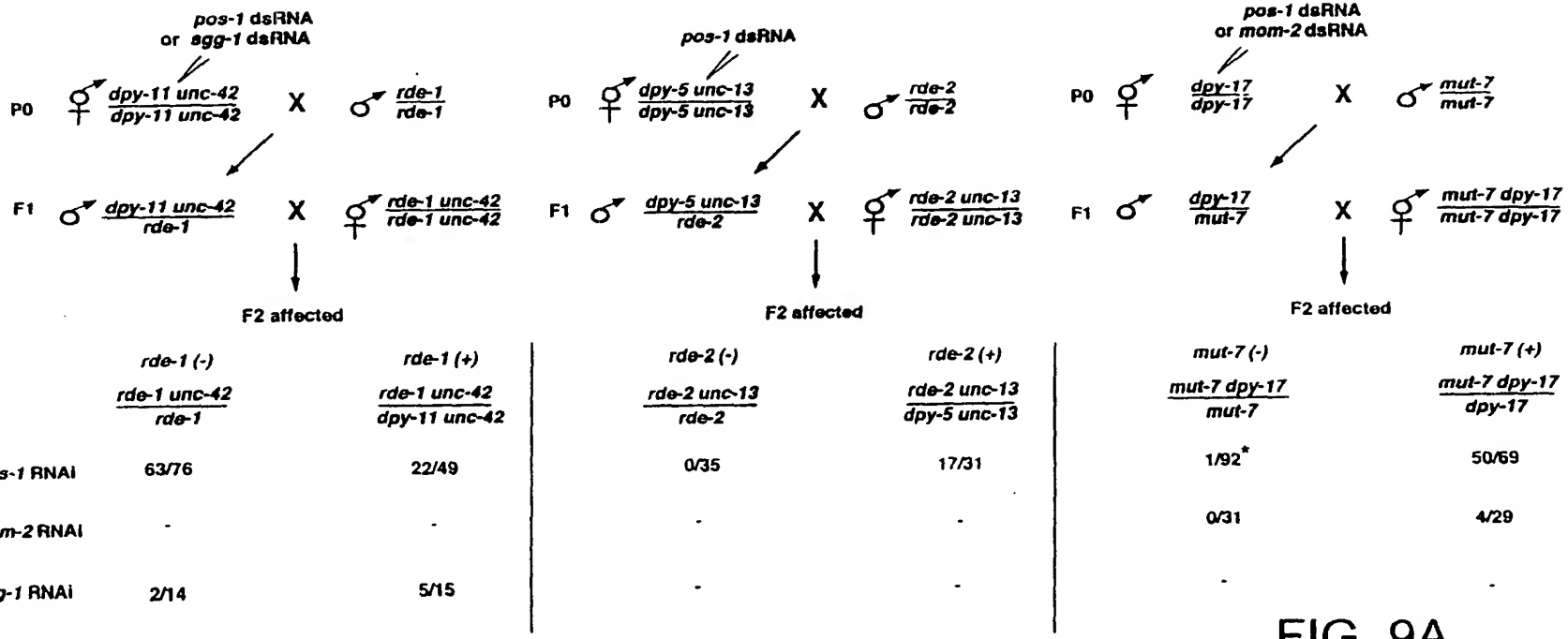

P0
Injected F1
rde-1 unc-42
+
pos-1 dsRNA
rde-1 unc-42
rde-1 unc-42
0/37
rde-4 unc-69
+
rde-4 unc-69
rde-4 unc-69
0/37

FIG. 8B



	rde-1 (-) rde-1 unc-42 / rde-1	rde-1 (+) rde-1 unc-42 / dpy-11 unc-42	rde-2 (-) rde-2 unc-13 / rde-2	rde-2 (+) rde-2 unc-13 / dpy-5 unc-13	mut-7 (-) mut-7 dpy-17 / mut-7	mut-7 (+) mut-7 dpy-17 / dpy-17
pos-1 RNAi	63/76	22/49	0/35	17/31	1/92*	50/69
mom-2 RNAi	-	-	-	-	0/31	4/29
sgg-1 RNAi	2/14	5/15	-	-	-	-

FIG. 9A

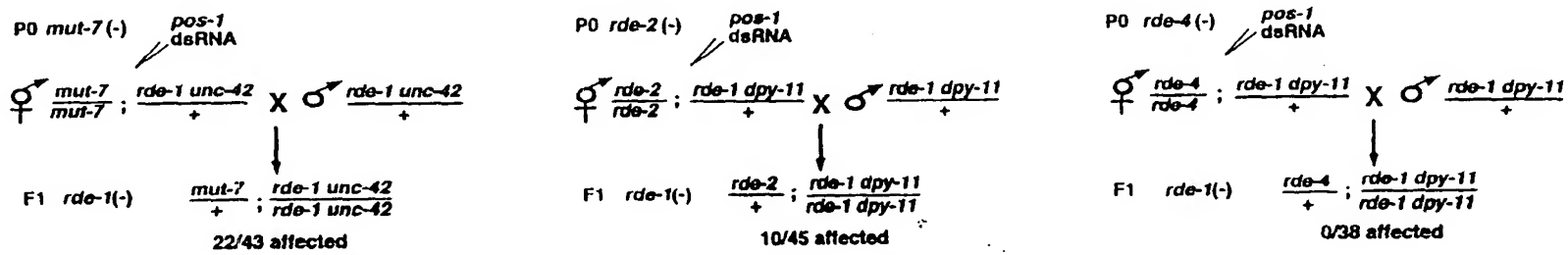


FIG. 9B

```
        10         20         30         40         50         60
ATGGATTTAACCAAACTAACGTTTGAAAGCGTTTTCGGTGGATCAGATGTTCCTATGAAG
M  D  L  T  K  L  T  F  E  S  V  F  G  G  S  D  V  P  M  K

        70         80         90        100        110        120
CCTTCCCGATCGGAGGATAACAAAACGCCAAGAAACAGAACAGATTTGGAGATGTTTCTG
P  S  R  S  E  D  N  K  T  P  R  N  R  T  D  L  E  M  F  L

       130        140        150        160        170        180
AAGAAAACTCCCCTCATGGTACTAGAAGAGGCTGCTAAGGCTGTCTATCAAAAGACGCCA
K  K  T  P  L  M  V  L  E  E  A  A  K  A  V  Y  Q  K  T  P

       190        200        210        220        230        240
ACTTGGGGCACTGTCGAACTTCCTGAAGGCTTCGAGATGACGTTGATTCTGAATGAAATT
T  W  G  T  V  E  L  P  E  G  F  E  M  T  L  I  L  N  E  I

       250        260        270        280        290        300
ACTGTAAAAGGCCAGGCAACAAGCAAGAAAGCTGCGAGACAAAAGGCTGCTGTTGAATAT
T  V  K  G  Q  A  T  S  K  K  A  A  R  Q  K  A  A  V  E  Y

       310        320        330        340        350        360
TTACGCAAGGTTGTGGAGAAAGGAAAGCACGAAATCTTTTTCATTCCTGGAACAACCAAA
L  R  K  V  V  E  K  G  K  H  E  I  F  F  I  P  G  T  T  K

       370        380        390        400        410        420
GAAGAAGCTCTTTCGAATATTGATCAAATATCGGATAAGGCTGAGGAATTGAAACGATCA
E  E  A  L  S  N  I  D  Q  I  S  D  K  A  E  E  L  K  R  S

       430        440        450        460        470        480
ACTTCAGATGCTGTTCAGGATAACGATAACGATGATTCGATTCCTACAAGTGCTGAATTT
T  S  D  A  V  Q  D  N  D  N  D  D  S  I  P  T  S  A  E  F

       490        500        510        520        530        540
CCACCTGGTATTTCGCCAACCGAGAATTGGGTCGGAAAGTTGCAGGAAAAATCTCAAAAA
P  P  G  I  S  P  T  E  N  W  V  G  K  L  Q  E  K  S  Q  K

       550        560        570        580        590        600
AGCAAGCTGCAAGCCCCAATCTATGAAGATTCCAAGAATGAGAGAACCGAGCGTTTCTTG
S  K  L  Q  A  P  I  Y  E  D  S  K  N  E  R  T  E  R  F  L

       610        620        630        640        650        660
GTTATATGCACGATGTGCAATCAAAAAACCAGAGGAATCAGAAGTAAGAAGAAGGACGCA
V  I  C  T  M  C  N  Q  K  T  R  G  I  R  S  K  K  K  D  A

       670        680        690        700        710        720
AAGAATCTTGCAGCATGGTTGATGTGGAAAGCGTTGGAAGACGGTATCGAATCTCTGGAA
K  N  L  A  A  W  L  M  W  K  A  L  E  D  G  I  E  S  L  E

       730        740        750        760        770        780
TCATATGATATGGTTGATGTGATTGAAAATTTGGAAGAAGCTGAACATTTACTCGAAATT
S  Y  D  M  V  D  V  I  E  N  L  E  E  A  E  H  L  L  E  I
```

FIG. 10A

```
      790       800       810       820       830       840
CAGGATCAAGCATCCAAGATTAAAGACAAGCATTCCGCACTGATTGATATACTCTCGGAC
Q  D  Q  A  S  K  I  K  D  K  H  S  A  L  I  D  I  L  S  D

      850       860       870       880       890       900
AAGAAAAGATTTTCAGACTACAGCATGGATTTCAACGTATTATCAGTGAGCACAATGGGA
K  K  R  F  S  D  Y  S  M  D  F  N  V  L  S  V  S  T  M  G

      910       920       930       940       950       960
ATACATCAGGTGCTATTGGAAATCTCGTTCCGGCGTCTAGTTTCTCCAGACCCCGACGAT
I  H  Q  V  L  L  E  I  S  F  R  R  L  V  S  P  D  P  D  D

      970       980       990      1000      1010      1020
TTGGAAATGGGAGCAGAACACACCCAGACTGAAGAAATTATGAAGGCTACTGCCGAGAAG
L  E  M  G  A  E  H  T  Q  T  E  E  I  M  K  A  T  A  E  K

     1030      1040      1050      1060      1070      1080
GAAAAGCTACGGAAGAAGAATATGCCAGATTCCGGGCCGCTAGTGTTTGCTGGACATGGT
E  K  L  R  K  K  N  M  P  D  S  G  P  L  V  F  A  G  H  G

     1090      1100      1110      1120      1130      1140
TCATCGGCGGAAGAGGCTAAACAGTGTGCTTGTAAATCGGCGATTATCCATTTCAACACC
S  S  A  E  E  A  K  Q  C  A  C  K  S  A  I  I  H  F  N  T

     1150      1160      1170      1180      1190      1200
TATGATTTCACGGATTGAAAATATTATTGCGTATTCCTGAAAAATGAAGCGTCTGAATGA
Y  D  F  T  D  *  K  Y  Y  C  V  F  L  K  N  E  A  S  E  *

     1210      1220      1230
TTATAAAAAAAAAAAAAAAAAA                    (SEQ ID NO:4)
L  *  K  K  K  K  K                        (SEQ ID NO:5)
```

FIG. 10B

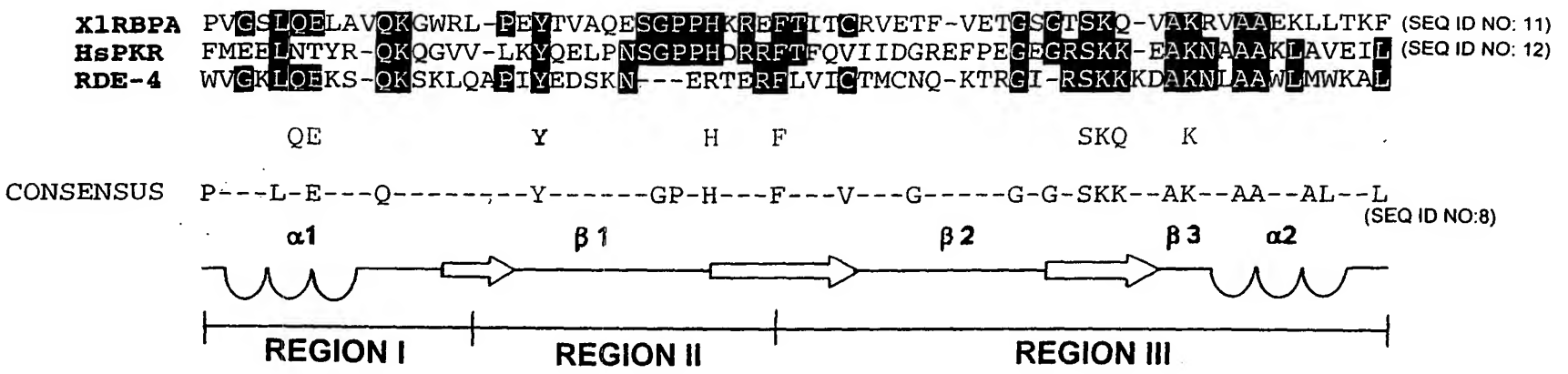

XlRBPA PVCSLQELAVQKGWRL-PEYTVAQESGPPHKREFTITCRVETF-VETGSGTSKQ-VAKRVAAEKLLTKF (SEQ ID NO: 11)
HsPKR FMEELNTYR-QKQGVV-LKYQELPNSGPPHDRRFTFQVIIDGREFPEGEGRSKK-EAKNAAAKLAVEIL (SEQ ID NO: 12)
RDE-4 WVGKLQEKS-QKSKLQAPIYEDSKN---ERTERFLVICTMCNQ-KTRGI-RSKKKDAKNLAAWLMWKAL
QE Y H F SKQ K
CONSENSUS P---L-E---Q------,--Y------GP-H---F---V---G-----G-G-SKK--AK--AA--AL--L
(SEQ ID NO:8)
α1 β1 β2 β3 α2
REGION I
REGION II
REGION III

FIG. 11

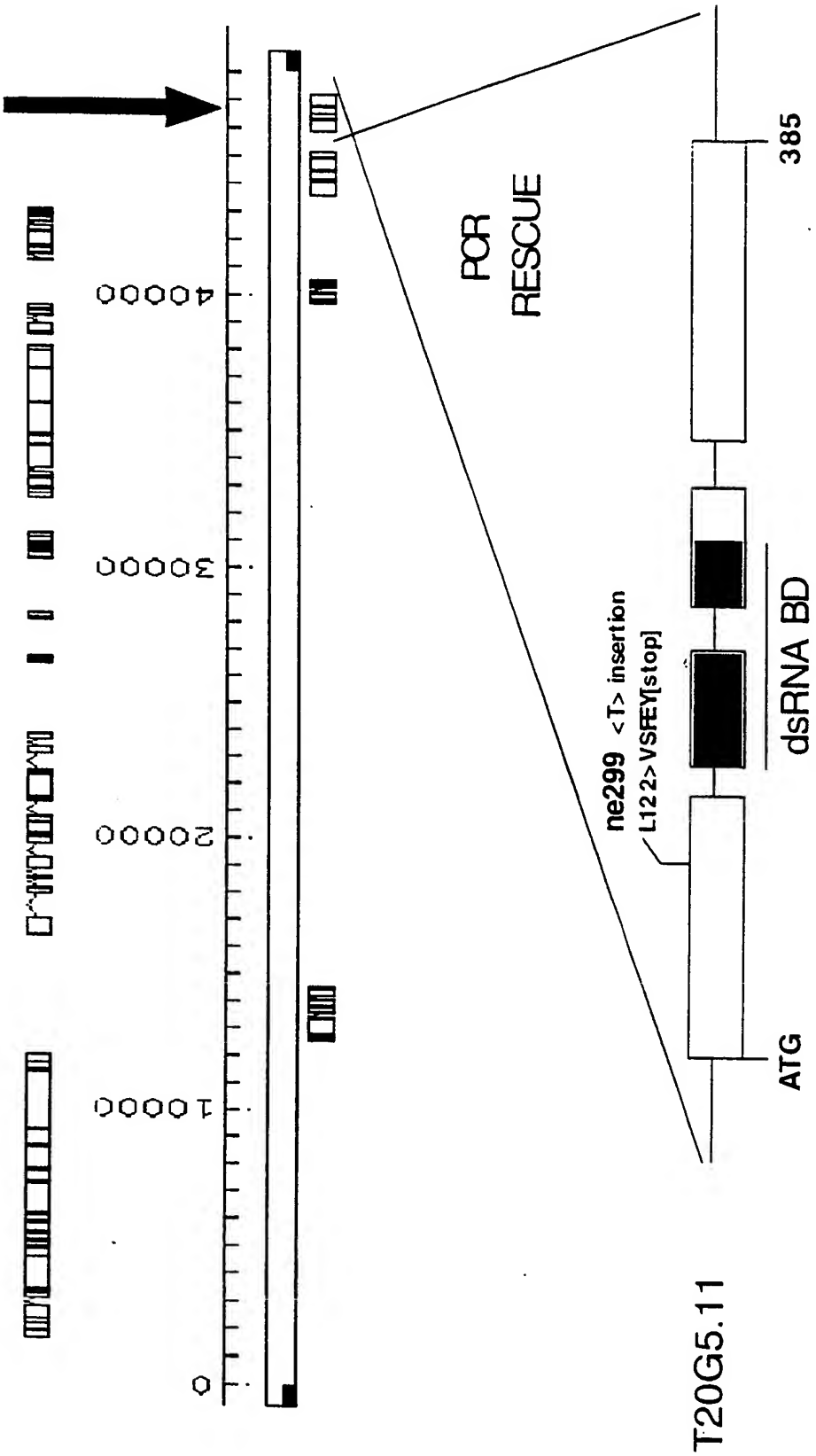



FIG. 12 Rescue of *rde-4*: